JACC: BASIC TO TRANSLATIONAL SCIENCE VOL. 5, NO. 7, 2020
© 2020 THE AUTHORS. PUBLISHED BY ELSEVIER ON BEHALF OF THE AMERICAN COLLEGE OF CARDIOLOGY FOUNDATION. THIS IS AN OPEN ACCESS ARTICLE UNDER THE CC BY-NC-ND LICENSE (http://creativecommons.org/licenses/by-nc-nd/4.0/).

EDITORIAL COMMENT

Neprilysin in HFrEF

Differential Regulation of Tissue Versus Circulating Neprilysin*

Horng H. Chen, MB, BCH, John C. Burnett, JR, MD

Neprilysin (NEP) is a ubiquitous zinc-dependent membrane metallo-endopeptidase (EC 3.4.24.11) that is expressed in multiple organs such as the kidneys, lungs, endothelial cells, vascular smooth muscle cells, cardiac cells, fibroblasts, neutrophils, adipocytes, testes, and brain. NEP cleaves numerous vasoactive peptides, such as bradykinin, substance P, natriuretic peptides, adrenomedullin, angiotensins, and endothelins (1). An alternative processing form of nonmembrane-associated soluble and/or circulating NEP that exists in plasma, urine, and cerebrospinal fluid (2). Studies have demonstrated that circulating NEP has an enzymatic activity similar to membrane-bound NEP (3). Alterations of circulating NEP concentrations and NEP activity were found in various diseases, such as cardiovascular disease, metabolic syndrome, lung disease, chronic rheumatoid disease, and Alzheimer's disease. In heart failure (HF), plasma concentrations of NEP were found to be a risk factor for cardiovascular death in stable patients and were adversely associated with outcome in acute HF.

Over the past 3 decades, NEP inhibition has been the target in cardiovascular therapeutics. Omapatrilat, a NEP inhibitor combined with an angiotensin-converting enzyme inhibitor (ACEI), was the first to be tested in both hypertension and heart failure phase III studies. However, despite demonstrating beneficial clinical effects, the clinical development was abandoned due to increased risk of incidence of angioedema. In 2015, a combination of a NEP inhibitor (sacubitril) and an angiotensin II type 1 receptor blocker (valsartan), LCZ 696, was approved by the Food and Drug Administration for the management of chronic HF with reduced ejection fraction (HFrEF) (2). The PARADIGM-HF (Prospective Comparison of ARNI With ACEI to Determine Impact on Global Mortality and Morbidity in Heart Failure) trial demonstrated significantly beneficial clinical endpoints with no excess adverse effects with LCZ696 compared with enalapril alone in patients with chronic HFrEF. Although it has been established that the renin-angiotensin-aldosterone system is activated in HFrEF, the pathophysiological relevance of membrane-bound tissue NEP and soluble and/or circulating NEP activity in HFrEF compared with the healthy state remains poorly defined.

In this issue of *JACC: Basic to Translational Science*, Pavo et al. (4) investigated differential NEP expression, NEP protein concentrations, and enzymatic NEP activity of various tissues and the relationship between tissue NEP concentrations and activity with circulating NEP status in a porcine myocardial infarction model of chronic HFrEF. The investigators confirmed findings from previous studies that the NEP content and activity of the kidneys were 20 to 100 times higher than that of any other organ. They determined that overall tissue NEP expression was downregulated and translated into reduced protein concentrations and activity in animals with HF compared with healthy control animals. Although tissue NEP activity correlated strongly with expression and protein concentrations ($r = 0.79$; $p < 0.001$ and $r = 0.85$; $p < 0.001$), plasma NEP concentrations

*Editorials published in *JACC: Basic to Translational Science* reflect the views of the authors and do not necessarily represent the views of *JACC: Basic to Translational Science* or the American College of Cardiology.

From the Department of Cardiovascular Diseases, Cardiorenal Research Laboratory, Mayo Clinic, Rochester, Minnesota. Dr. Chen was supported by grants from the National Institutes of Health (NHLBI R01HL84155 and R01-HL136440). Dr. Burnett was supported by the National Institutes of Health (RO1 HL134668). Drs. Chen and Burnett hold patents for designer peptides and are co-founders of Zumbro Discovery.

The authors attest they are in compliance with human studies committees and animal welfare regulations of the authors' institutions and Food and Drug Administration guidelines, including patient consent where appropriate. For more information, visit the *JACC: Basic to Translational Science* author instructions page.

ISSN 2452-302X https://doi.org/10.1016/j.jacbts.2020.05.010

JACC: BASIC TO TRANSLATIONAL SCIENCE VOL. 5, NO. 7, 2020
JULY 2020:727–9

and activity were not significantly correlated to their tissue equivalents.

The expression of membrane bound NEP, protein concentrations, and enzymatic activity of various tissues in HFrEF is controversial. Knecht et al. (1) determined that renal NEP activity was increased in rodent models of heart failure (e.g., aortocaval shunt, aortic banding, and infarction). Similarly, previous studies have demonstrated in human left ventricle biopsies that myocardial NEP expression and activity were increased in humans with aortic stenosis and dilated cardiomyopathy. In contrast, the current study by Pavo et al. (4) reported that NEP activity was decreased in the porcine infarction model. Furthermore, both Abassi et al. (5) and Huang et al. (6) reported that neither renal NEP expression nor activity were increased in rodent models of HF.

One explanation for this discrepancy might be the different severity of HF in the different studies. Abassi et al. (5) demonstrated that pulmonary NEP expression and activity decreased by 45% in rats with severe HF compared with those with mild compensated HF. Therefore, it could be hypothesized that in mild compensated HF, NEP activity is maintained or upregulated; however, with progression to severe HF, NEP is downregulated. Pre-clinical studies have demonstrated enhanced effects of NEP inhibition in mild compensated HF but not in severe HF, which supports this paradigm (7). The differential regulation of NEP in mild HFrEF versus severe HFrEF may have important clinical implications with the use of an angiotensin receptor and neprilysin inhibitor (ARNI) in HFrEF. Most patients (95%) in the PARADIGM-HF trial were in New York Heart Association (NYHA) functional classes II to III HF status. Hence, the efficacy of LCZ 696 in patients in NYHA functional class IV has not been well defined and is currently being evaluated in the LIFE (Entresto [LCZ696] In Advanced Heart Failure; NCT02816736) study sponsored by National Heart, Lung, and Blood Institute Heart Failure Clinical Research Network.

Recent studies have suggested that circulating NEP has potential as a biomarker for the prognosis of cardiovascular death and hospital admission for patients with acute and chronic HF. Are circulating NEP levels and NEP activity indicators of the tissue activation and/or inhibition of the NEP system in HF? Previously, there were no data regarding this question. However, the current study by Pavo et al. (4) demonstrated that circulating NEP concentrations and activity were not significantly correlated with their tissue equivalents. The fact that circulating NEP concentration and activity was not associated with tissue NEP does not preclude it as a biomarker in HF nor as having degradative activity. In studies of pro–B-type natriuretic peptide (BNP) and mature BNP in plasma from normal subjects and subjects with HF, incubation of BNP in HF plasma was characterized by a delayed degradation compared with normal individuals, which suggested that plasma NEP enzymatic activity might be functional (3). Furthermore, Nougue et al. (2) reported that NEP activity in the plasma was reduced by sacubatril/valsartan therapy (2). Nonetheless, studies are warranted to understand the regulation and function of circulating NEP.

Pavo et al. (4) also reported that there was no correlation between tissue and circulating NEP concentration and activity with natriuretic peptide levels. This was consistent with previous studies in patients with HF and the general population. This may be due to the fact that the natriuretic peptides are also cleared by the clearance receptor and metabolized other enzymes, such as the insulin degrading enzyme and dipeptidyl-peptidase IV. Because Reginauld et al. (8) reported that 25% of patients with HF are deficient in atrial natriuretic peptides (ANPs), it may be important to understand fully if ANP, which is increased by sacubatril/valsartan in contrast to BNP, elevation or deficiency is related to reduced and/or increased circulating NEP.

In summary, the role of NEP as a regulator of numerous vasoactive peptides has been established in HFrEF. Insights from the current and previous investigations suggest that the regulation of NEP in HFrEF is dynamic and may alter with the progression of the disease. Although recent studies suggest that circulating NEP may have a potential role as a biomarker with prognostic value in HFrEF, the regulation and function of circulating NEP is not well defined and needs to be addressed. From a therapeutic standpoint, the clinical efficacy of ARNI in patients with HFrEF in NYHA functional classes II and/or III has been established. The results of the LIFE study will provide more information on the efficacy of ARNI in patients with advanced HF.

ADDRESS FOR CORRESPONDENCE: Dr. Horng H. Chen, Department of Cardiovascular Diseases, Cardiorenal Research Laboratory, Mayo Clinic, 200 First Street Southwest, Rochester, Minnesota 55905. E-mail: chen.horng@mayo.edu.

REFERENCES

1. Knecht M, Pagel I, Langenickel T, et al. Increased expression of renal neutral endopeptidase in severe heart failure. Life Sci 2002;71:2701–12.

2. Nougue H, Pezel T, Picard F, et al. Effects of sacubitril/valsartan on neprilysin targets and the metabolism of natriuretic peptides in chronic heart failure: a mechanistic clinical study. Eur J Heart Fail 2019;21:598–605.

3. Huntley BK, Sandberg SM, Heublein DM, Sangaralingham SJ, Burnett JC JR., Ichiki T. Pro-B-type natriuretic peptide-1-108 processing and degradation in human heart failure. Circ Heart Fail 2015;8:89–97.

4. Pavo IJ, Pavo N, Kastner N, et al. Heart failure with reduced ejection fraction is characterized by systemic NEP down-regulation. J Am Coll Cardiol Basic Trans Science 2020;5:727–9.

5. Abassi ZA, Kotob S, Golomb E, Pieruzzi F, Keiser HR. Pulmonary and renal neutral endopeptidase EC 3.4.24.11 in rats with experimental heart failure. Hypertension 1995;25:1178–84.

6. Huang H, Arnal JF, Llorens-Cortes C, et al. Discrepancy between plasma and lung angiotensin-converting enzyme activity in experimental congestive heart failure. A novel aspect of endothelium dysfunction. Circ Res 1994;75:454–61.

7. Chen HH, Schirger JA, Chau WL, et al. Renal response to acute neutral endopeptidase inhibition in mild and severe experimental heart failure. Circulation 1999;100:2443–8.

8. Reginauld SH, Cannone V, Iyer S, et al. Differential regulation of ANP and BNP in acute decompensated heart failure: deficiency of ANP. J Am Coll Cardiol HF 2019;7:891–8.

KEY WORDS heart failure, natriuretic peptides, neprilysin